## Remarks

The Office Action mailed October 15, 2002 has been received and reviewed. Claims 12, 31, 38, and 41 having been amended, the pending claims are claims 1-45. Reconsideration and withdrawal of the rejections are respectfully requested.

The specification has been amended to correct obvious errors in nomenclature. It would be obvious to one of skill in the art that dNTPs refer to deoxynucleotide triphosphates and/or deoxynucleoside triphosphates.

Claims 12, 38, and 41 have been amended, as described herein for the specification, to correct obvious errors in nomenclature. Claim 38 and 41 have also been amended to recite that "the dye inactivates the enzyme in the absence of the surfactant, and the surfactant inhibits such inactivation," which is supported, for example, by originally filed claim 1.

### The 35 U.S.C. §103 Rejection

The Examiner rejected claims 1, 5-16, 20, and 23 under 35 U.S.C. §103(a) as allegedly being unpatentable over U.S. Pat. No. 5,861,251 (Park et al.) in view of U.S. Pat. Nos. 6,242,235 B1 (Shultz et al.) and 5,721,123 (Hayes et al.). Applicants respectfully disagree.

Independent claim 1 (as well as independent claims 17, 18, and 19, which are not subject to this rejection) recites that a "dye inactivates the enzyme in the absence of the surfactant." Independent claim 20 (as well as independent claim 24, which is not subject to this rejection) recites the presence of a dye "under conditions that normally inactivate the enzyme." Applicants respectfully submit that none of the cited documents teach or suggest all the claim language including, for example, a composition in which a "dye inactivates the enzyme in the absence of the surfactant" (e.g., claims 1-19), or a method of stabilizing an enzyme in the presence of a dye "under conditions that normally inactivate the enzyme," wherein the method includes "combining an effective amount of a . . . surfactant . . . with the enzyme and the dye (e.g., claims 20-24).

> "To establish a *prima facie* case of obviousness, three basic criteria must be met. First, there must be some suggestion or motivation, either in the references themselves or in the knowledge generally available to one of ordinary skill in the art, to modify the reference or to combine reference teachings. Second, there must be a reasonable expectation of success. Finally, the prior art reference (or references when combined) must teach or suggest all the claim limitations. The teaching or suggestion to make the claimed combination and the reasonable expectation of success must both be found in the prior art and not based on applicant's disclosure."

M.P.E.P. §706.02(j). Applicants respectfully submit that the Examiner has failed to establish a *prima facie* case of obviousness.

Specifically, Park et al. disclose that a "reagent for PCR . . . is prepared by freeze-drying a conventional aqueous reaction mixture which consists of a reaction buffer, $MgCl_2$, dNTPs and a DNA polymerase" (column 3, lines 3-7). "The PCR reagent of the invention may further comprise a sedimenting agent or a water-soluble dye in the presence/absence of stabilizer" (column 3, lines 30-32). "It has been well known that: materials such as gelatin, bovine serum albumin (BSA), ammonium sulfate or Thesit etc., stabilize a DNA polymerase and dNTPs, and non-ionic surfactants such as NP40 and Tween 20 etc., improve the reactivity of the PCR mixture" (column 3, lines 15-20).

The Examiner asserted that Park et al. disclose a PCR reagent mixture containing a polymerase, a dye, and a nonionic surfactant. Although Park et al. disclose that a PCR reagent may include "a sedimenting agent or a water-soluble dye in the presence/absence of stabilizer" (column 3, lines 30-32), Applicants respectfully submit that Park et al. fail to explicitly disclose a PCR reagent mixture that includes a polymerase, *a dye, and a nonionic surfactant*. For example, PCR reagent mixtures that include a nonionic surfactant (e.g., Thesit) are explicitly disclosed at column 4, line 21 to column 5, line 18 (Examples 2-4), all of which are reagent mixtures that do not include a dye. Furthermore, Applicants respectfully submit that the only PCR reagent mixtures including a dye and explicitly disclosed by Park et al. are at column 6, lines 1-29

(Example 7), which are reagent mixtures that do not include a nonionic surfactant.

Shultz et al. disclose "methods and compositions for protein stabilization, particularly the stabilization of polymerases in aqueous solutions with cationic surfactants. The activity of polymerases in solution, either in storage buffers or reaction buffers, may be stabilized by the addition of non-ionic surfactants" (column 6, lines 38-43). However, Shultz et al. fail to teach or suggest, among other things, any methods or compositions that include a dye.

Hayes et al. disclose "methods and apparatus for changing the temperature of material in a vessel by exposing the vessel to electromagnetic radiation" (column 1, lines 5-8). Hayes et al. further disclose the use of "heat absorptive dyes . . . for enhancing the heating effect of the electromagnetic radiation" (column 3, lines 10-12). However, Hayes et al. fail to teach or suggest, among other things, any methods or apparatuses that include a surfactant.

Further, the Examiner asserted that "[t]he dye of Park et al and/or the heat absorptive dye of Hayes et al would have *inherently* reduced polymerase activity in the absence of the surfactant" (page 3, lines 18-20, of Office Action mailed October 15, 2002, emphasis added). Applicants respectfully submit that *inherency* may not be used to properly support an obviousness rejection under 35 U.S.C. §103: "[T]he prior art reference (or references when combined) *must teach or suggest all the claim limitations*" (M.P.E.P. §2143, emphasis added).

As described hereinabove, Applicants respectfully submit that Park et al. explicitly disclose PCR reagent mixtures that include a dye, but do not include a surfactant, at column 6, lines 1-29 (Example 7). "[W]ater soluble dyes . . . were added to the conventional PCR mixture of Example 1" (column 6, lines 6-9). Example 1 included BSA (column 4, line 11), a material known to "stabilize a DNA polymerase and dNTPs" (column 3, lines 16-18). Moreover, Park et al. report that "there was no decrease in the level of PCR, except for the case of methyl green-added reaction mixture. *This result validates that bromophenol blue, xylene cyanole, bromocresol red and cresol red can be efficiently added in the preparation of the lyophilized PCR mixtures*" (column 6, lines 16-21, emphasis added). Based on the remarks presented herein

above, Applicants respectfully submit that Park et al., do not disclose, and in fact teach away from, a composition in which a "dye inactivates the enzyme in the absence of the surfactant" (e.g., claims 1-19), or a method of stabilizing an enzyme in the presence of a dye "under conditions that normally inactivate the enzyme," wherein the method includes "combining an effective amount of a . . . surfactant . . . with the enzyme and the dye" (e.g., claims 20-24).

Applicants respectfully submit that Park et al. in view of Shultz et al. and Hayes et al. fail to teach or suggest the presently claimed invention. Furthermore, Park et al., Shultz et al., and Hayes et al. provide no suggestion or motivation for one of skill in the art to modify or to combine their teachings to arrive at the present invention with a reasonable expectation of success.

The Examiner rejected claims 2-4, 17-19, 21, and 24 under 35 U.S.C. §103(a) as allegedly being unpatentable over U.S. Pat. No. 5,861,251 (Park et al.) in view of U.S. Pat. Nos. 6,242,235 B1 (Shultz et al.) and 5,721,123 (Hayes et al.), and further in view of U.S. Pat. No. 5,919,630 (Nadeau et al.). Applicants respectfully disagree.

The deficiencies of Park et al. in view of Shultz et al. and Hayes et al., as applied to claims 1-24, have been discussed herein above.

Nadeau et al. disclose "methods for detecting nucleic acid target sequences" (column 1, lines 9-10). However, Nadeau et al. fail to disclose or suggest, among other things, any methods that include a surfactant. Thus, Nadeau et al. provide nothing to correct the deficiencies of Park et al. in view of Shultz et al. and Hayes et al. Furthermore, Applicants respectfully submit that one cannot simply engage in a hindsight reconstruction of the claimed invention, using Applicants' structure as a template and selecting elements from documents to fill the gaps.

Based on the remarks presented herein above, Applicants respectfully submit that claims 2-4, 17-19, 21, and 24 are patentable over Park et al. in view of Shultz et al. and Hayes et al., and further in view of Nadeau et al.

**Obviousness-Type Double Patenting Rejection**

Claims 1-24 were provisionally rejected under the judicially created doctrine of obviousness-type double patenting as being unpatentable over claims 1-45 of copending Application No. 09/841,272. Upon an indication of otherwise allowable subject matter and in the event this rejection is maintained, Applicants will provide an appropriate response.

If the provisional obviousness-type double patenting rejection is the only rejection remaining in this application, the Examiner is requested to withdraw the rejection and permit the application to issue as a patent, thereby allowing any such issues to be resolved in the issuance of the other application. M.P.E.P. §804 affirmatively directs the Examiner to withdraw the provisional obviousness-type double patenting rejection under such circumstances.

Reconsideration and withdrawal of the provisional obviousness-type double patenting rejection are respectfully requested.

**Request for Rejoinder**

Claims 24-45 have been withdrawn by the Examiner from further consideration as being drawn to a non-elected invention. Reconsideration and withdrawal of the restriction requirement is respectfully requested.

In the event the Examiner maintains the restriction requirement, rejoinder of the non-elected claims is respectfully requested. Notably, all the independent claims that have been withdrawn from further consideration (e.g., claims 25, 31, 41, and 45) are method claims that recite all the language of the composition of, for example, independent claim 1. *See* 1184 O.G. 86 citing *In re Ochiai*, 37 USPQ2d 1127 (Fed. Cir. 1995) and *In re Brouwer*, 37 USPQ2d 1663 (Fed. Cir. 1996).

**Information Disclosure Statement**

Applicants submitted an Information Disclosure Statement mailed on February 28, 2002. Pursuant to the provisions of M.P.E.P. §609, Applicants further request that a copy of the 1449 form(s), marked as being considered and initialed by the Examiner, be returned with the next Official Communication. For the convenience of the Examiner, attached herewith are copies of the 1449 forms.

## Summary

It is respectfully submitted that all the pending claims are in condition for allowance and notification to that effect is respectfully requested. The Examiner is invited to contact Applicants' Representatives, at the below-listed telephone number, if it is believed that prosecution of this application may be assisted thereby.

Respectfully submitted for
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CERTIFICATE UNDER 37 CFR §1.8:
The undersigned hereby certifies that this paper is being transmitted by facsimile in accordance with 37 CFR §1.6(d) to the Patent and Trademark Office, addressed to Assistant Commissioner for Patents, Washington, D.C. 20231, on this 15 day of January, 2003, at 10:37 a.m. (Central Time).

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**APPENDIX A - SPECIFICATION/CLAIM AMENDMENTS INCLUDING NOTATIONS TO INDICATE CHANGES MADE**
**Serial No.: 09/841,264**
**Docket No.: 56286US003**

Amendments to the following are indicated by underlining what has been added and bracketing what has been deleted. Additionally, all amendments have been marked in bold typeface.

**In the Specification**

The paragraph beginning at page 14, line 28, has been amended as follows:

The methods described herein can be used in a variety of different processes requiring thermal cycling of samples contained in the process chambers of the devices. Examples of some such processes involve chemical reactions of samples, e.g., nucleic acid amplification. For example, samples may be mixed with a polynucleotide, a polymerase (such as *Taq* polymerase), **[nucleoside]** triphosphates **(e.g., dNTPs)**, a first primer hybridizable with the sample polynucleotide, and a second primer hybridizable with a sequence complementary to the polynucleotide. Some or all of the required reagents may be present in the device as manufactured, they may be loaded into the process chambers after manufacture of the device, they may be loaded in the process chambers just before introduction of the sample, or they may be mixed with sample before loading into the process chambers.

The paragraph beginning at page 15, line 26, has been amended as follows:

Of the potential uses for the devices and methods of the present invention, PCR is one important such use, although it should be understood that the present invention is not limited to PCR amplification. PCR allows for analysis of extremely small amounts of target nucleic acid (e.g., DNA) using an excess of two oligonucleotide primers that are capable of flanking the region of the denatured molecule to be amplified and extending the nucleic acid molecule by nucleotide addition from the primers by the action of a polymerase enzyme (such as Taq DNA polymerase) in the presence of free **[deoxynucleoside triphosphates (dNTPs)]dNTPs (also**

**referred to herein as deoxynucleotide triphosphates and/or deoxynucleoside triphosphates),** resulting in a double replication of the starting target nucleic acid molecule. The nucleic acid molecules are again thermally treated to denature, and the process is repeated to form PCR amplification products (also referred to as PCR amplicons).

## In the Claims

For convenience, all pending claims are shown below.

1. A composition comprising an enzyme, a dye, and an effective amount of a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof, wherein the dye inactivates the enzyme in the absence of the surfactant, and the surfactant inhibits such inactivation.

2. The composition of claim 1 wherein the dye is selected from the group of a near-IR dye, a uv/visible dye, a fluorescent dye, and a mixture thereof.

3. The composition of claim 2 wherein the dye is a near-IR dye.

4. The composition of claim 3 wherein the near-IR dye is a diimminium dye or a cyanine dye.

5. The composition of claim 1 wherein the enzyme is a polymerase or a ligase.

6. The composition of claim 1 wherein the nonionic surfactant is selected from the group of esters of fatty acids and polyhydric alcohols, fatty acid alkanolamides, ethoxylated fatty acids, ethoxylated aliphatic acids, ethoxylated fatty alcohols, ethoxylated aliphatic alcohols,

ethoxylated sorbitol fatty acid esters, ethoxylated glycerides, ethoxylated block copolymers with EDTA, ethoxylated cyclic ether adducts, ethoxylated amide and imidazoline adducts, ethoxylated amine adducts, ethoxylated mercaptan adducts, ethoxylated condensates with alkyl phenols, ethoxylated nitrogen-based hydrophobes, ethoxylated polyoxypropylenes, polymeric silicones, fluorinated surfactants, polymerizable surfactants, and mixtures thereof.

7. The composition of claim 1 wherein the zwitterionic surfactant is selected from the group of alkylamido betaines and amine oxides thereof, alkyl betaines and amine oxides thereof, sulfo betaines, hydroxy sulfo betaines, amphoglycinates, amphopropionates, balanced amphopolycarboxyglycinates, and alkyl polyaminoglycinates, and mixtures thereof.

8. The composition of claim 1 wherein the dye is present at a concentration of at least about 0.005 mg/mL.

9. The composition of claim 1 wherein the effective amount of surfactant is at least about 0.5 wt-%.

10. The composition of claim 9 wherein the effective amount of surfactant is no greater than about 20 wt-%.

11. The composition of claim 1 further comprising a buffer.

12. **(Amended)** The composition of claim 1 further comprising a **[dinucleotide]** triphosphate.

13. The composition of claim 1 further comprising a reference dye.

14. The composition of claim 1 further comprising an antioxidant.

15. The composition of claim 14 wherein the dye is capable of optical degradation.

16. The composition of claim 1 wherein the surfactant is an antioxidant.

17. A composition comprising a polymerase enzyme, a near-IR dye, and an effective amount of a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof, wherein the near-IR dye inactivates the enzyme in the absence of the surfactant, and the surfactant inhibits the inactivation.

18. A composition comprising:

a polymerase enzyme;

a near-IR dye selected from the group of a diimminium dye, a cyanine dye, and a mixture thereof; and

an effective amount of a nonionic surfactant;

wherein the near-IR dye inactivates the enzyme in the absence of the surfactant, and the surfactant inhibits the inactivation.

19. A composition comprising:

a polymerase enzyme;

a near-IR dye selected from the group of a diimminium dye, a cyanine dye, and a mixture thereof; and

an effective amount of a nonionic surfactant selected from the group of esters of fatty acids and polyhydric alcohols, fatty acid alkanolamides, ethoxylated fatty acids, ethoxylated

aliphatic acids, ethoxylated fatty alcohols, ethoxylated aliphatic alcohols, ethoxylated sorbitol fatty acid esters, ethoxylated glycerides, ethoxylated block copolymers with EDTA, ethoxylated cyclic ether adducts, ethoxylated amide and imidazoline adducts, ethoxylated amine adducts, ethoxylated mercaptan adducts, ethoxylated condensates with alkyl phenols, ethoxylated nitrogen-based hydrophobes, ethoxylated polyoxypropylenes, polymeric silicones, fluorinated surfactants, polymerizable surfactants, and mixtures thereof;

wherein the near-IR dye inactivates the enzyme in the absence of the surfactant, and the surfactant inhibits the inactivation.

20. A method of stabilizing an enzyme in a fluid sample in the presence of a dye under conditions that normally inactivate the enzyme, the method comprising combining an effective amount of a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof, with the enzyme and the dye, wherein the surfactant inhibits inactivation of the enzyme.

21. The method of claim 20 wherein the dye is selected from the group of a near-IR dye, a uv/visible dye, a fluorescent dye, and a mixture thereof.

22. The method of claim 21 wherein the enzyme is a polymerase or a ligase.

23. The method of claim 20 wherein the surfactant is a nonionic surfactant selected from the group of esters of fatty acids and polyhydric alcohols, fatty acid alkanolamides, ethoxylated fatty acids, ethoxylated aliphatic acids, ethoxylated fatty alcohols, ethoxylated aliphatic alcohols, ethoxylated sorbitol fatty acid esters, ethoxylated glycerides, ethoxylated block copolymers with EDTA, ethoxylated cyclic ether adducts, ethoxylated amide and imidazoline adducts, ethoxylated amine adducts, ethoxylated mercaptan adducts, ethoxylated

condensates with alkyl phenols, ethoxylated nitrogen-based hydrophobes, ethoxylated polyoxypropylenes, polymeric silicones, fluorinated surfactants, polymerizable surfactants, and mixtures thereof.

24. A method of stabilizing a polymerase enzyme in solution in the presence of a near-IR dye under conditions that normally inactivate the enzyme, the method comprising combining an effective amount of a nonionic surfactant with the enzyme and the dye, wherein the surfactant inhibits inactivation of the enzyme.

25. A method of conducting a thermal process, the method comprising:

providing a sample mixture comprising a biological material, an enzyme, an effective amount of a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof, and a dye at a first temperature; and

directly heating the sample mixture to a second temperature higher than the first temperature;

wherein the dye inactivates the enzyme in the absence of the surfactant and the surfactant inhibits the inactivation.

26. The method of claim 25 further comprising cooling the sample mixture and directly reheating the sample mixture in a thermal cycling process.

27. The method of claim 26 wherein the thermal cycling process comprises at least about 25 cycles.

28. The method of claim 27 wherein the first temperature is within a range of about 0°C to about 50°C.

29. The method of claim 27 wherein the second temperature is within a range of about 50°C to about 95°C.

30. The method of claim 27 wherein the thermal cycling process comprises heating between a temperature of about 50°C and about 95°C.

31. **(Amended)** A method of conducting a thermal cycling process, the method comprising:

providing a device comprising at least one process chamber that defines a volume for containing a sample mixture comprising a biological material, an enzyme, a dye, and **an effective amount of** a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof;

delivering electromagnetic energy to the process chamber to raise the temperature of the sample material in the process chamber, wherein the dye converts the electromagnetic energy into thermal energy;

wherein **the dye inactivates the enzyme in the absence of the surfactant, and** the surfactant inhibits **such inactivation[interaction between the enzyme and the dye]**.

32. The method of claim 31 wherein the dye is a near-IR dye.

33. The method of claim 31 wherein the surfactant is a nonionic surfactant selected from the group of esters of fatty acids and polyhydric alcohols, fatty acid alkanolamides, ethoxylated fatty acids, ethoxylated aliphatic acids, ethoxylated fatty alcohols, ethoxylated aliphatic alcohols, ethoxylated sorbitol fatty acid esters, ethoxylated glycerides, ethoxylated block copolymers with EDTA, ethoxylated cyclic ether adducts, ethoxylated amide and

imidazoline adducts, ethoxylated amine adducts, ethoxylated mercaptan adducts, ethoxylated condensates with alkyl phenols, ethoxylated nitrogen-based hydrophobes, ethoxylated polyoxypropylenes, polymeric silicones, fluorinated surfactants, polymerizable surfactants, and mixtures thereof.

34. The method of claim 31 wherein the sample mixture further comprises an antioxidant.

35. The method of claim 31 wherein the surfactant is present in an amount of at least about 0.5 wt-%.

36. The method of claim 35 wherein the surfactant is present in an amount of no greater than about 20 wt-%.

37. The method of claim 31 wherein the sample mixture further comprises a buffer.

38. **(Amended)** The method of claim 31 wherein the sample mixture further comprises a **[dinucleotide]** triphosphate.

39. The method of claim 31 wherein the sample mixture further comprises a reference dye.

40. The method of claim 31 wherein the enzyme is a polymerase or a ligase.

41. **(Amended)** A method of conducting a thermal cycling process comprising:

providing a device comprising at least one process chamber that defines a volume

for containing a sample mixture comprising a biological material, a polymerase enzyme, a near-IR dye, **an effective amount of** a nonionic surfactant, and a **[dinucleotide]** triphosphate;

delivering electromagnetic energy to the process chamber to raise the temperature of the sample material in the process chamber, wherein the dye converts the electromagnetic energy into thermal energy;

wherein **the dye inactivates the enzyme in the absence of the surfactant, and** the surfactant inhibits **such inactivation[interaction between the enzyme and the dye]**.

42. The method of claim 41 further comprising cooling the sample mixture and reheating the sample mixture in a thermal cycling process.

43. The method of claim 42 wherein the thermal cycling process comprises at least about 25 cycles.

44. The method of claim 42 wherein the thermal cycling process comprises heating between a temperature of about 50°C and about 95°C.

45. A method of denaturing hydrogen-bonded molecules, the method comprising:

providing a sample mixture comprising hydrogen-bonded molecules, an enzyme, an effective amount of a surfactant selected from the group of a nonionic surfactant, a zwitterionic surfactant, and a mixture thereof, and a dye at a first temperature; and

directly heating the sample mixture to a second temperature higher than the first temperature effective to denature the hydrogen-bonded molecules;

wherein the dye inactivates the enzyme in the absence of the surfactant and the surfactant inhibits the inactivation.